

RESEARCH-SPECIFIC STANDARD OPERATING PROCEDURE (SOP)

**PROCEDURE FOR WORK WITH MPTP
(1-methyl-4-phenyl-1,2,3,6-tetrahydropyridine CAS # 28289-54-5)
Or MPTP-Treated Animals**

HAZARD CLASS: TOXIC (NEUROTOXIN)

INVESTIGATOR'S NAME: Dr. Lester R. Drewes
PHONE NUMBER: 218-726-7925
EMERGENCY PHONE #: 726-7273 OR 911
WASTE DISPOSAL #: 726-6764

IACUC PROTOCOL # 0807A40601
IBC PROTOCOL #
APPROVAL DATE:
EFFECTIVE DATE:

1. SCOPE

To establish procedures for safe handling, transport and storage and disposal of MPTP ([1-methyl-4-phenyl-1,2,3,6-tetrahydropyridine](#), CAS # 28289-54-5), solutions of MPTP or MPTP contaminated materials or specimen.

MPTP is a potent neurotoxin known to produce severe irreversible brain damage similar to advanced Parkinson's disease. Unsafe handling of MPTP or MPTP-treated animals, contaminated materials, or sharps may result in neurological damage to the researchers therefore its handling, storage use and disposal must follow a strict safety protocol.

Signs and symptoms of the on-set of MPTP-associated disease include slowness of movement, postural instability, rigidity and tremors.

This SOP addresses laboratory use and proper handling of MPTP under the assumption that all laboratory staff have had the [required safety training](#), and are in compliance with minimum laboratory safety requirements, as detailed in the latest version of the UMD Medical School-Specific [Laboratory Safety Plan](#). The SOP covers

- Pure MPTP or any solution of MPTP, regardless of concentration.
- MPTP Treated animals and specimen.
- All MPTP-contaminated waste or labware materials
- Any excretions from test animals that would reasonably be expected to contain traces of MPTP and/or its metabolites.

2. RESPONSIBILITY

All staff engaged in the use or handling of MPTP, or working within a laboratory using MPTP are therefore responsible for understanding all hazards-associated with using MPTP, as well as appropriate use of protective equipment (PPE).

The Principal Investigator (PI) or his/her associate is responsible for ensuring that all the staff have been trained in the use, storage and handling of MPTP and are familiar with

RESEARCH-SPECIFIC STANDARD OPERATING PROCEDURE (SOP)

this SOP, and that MPTP use is restricted to the specific staff members who have had prior training in proper use, handling and storage of chemicals. Each staff member's name must appear on the staff listing section of this document, and each staff member must initial by his/her name.

3. PROCEDURES

A. Training Requirements

- Prior to starting work with MPTP, the PI must provide training to his/her laboratory personnel specific to the hazards associated with working with MPTP, work area decontamination, and emergency response procedures.
- The Principal Investigator must provide his/her laboratory personnel a copy of this SOP as well as copies of MSDS of all chemicals to be used in conjunction with this research including MPTP
- The Principal Investigator must ensure that his/her laboratory personnel have completed initial laboratory safety training or refresher safety training within the last year.

Note: The UMD Environmental Health and Safety Office staff is available to assist in the education and/or training of personnel concerning safe work practices. Also training may be taken online at: <http://www.d.umn.edu/ehso/safety/lsptrain.html>.

B. Occupational Safety and Health Program:

The PI must ensure that all laboratory staff associated with this research are all enrolled with the University Occupational Health and Safety Program (OHS), and must send personnel for medical evaluation to ensure they can safely take the **prescribed antidote** without any health risk.

Medical evaluation and clearance by OHS are also needed for any required use of respirators.

Antidote: [Selegiline HCL/deprenyl](#) (CAS # 14611-52-0), a [Monoamine oxidase inhibitors MAOI](#), is a potential antidote for MPTP exposure. For more information on Selegiline, see the following website:
<http://www.drugstore.com/pharmacy/drugindex/rxsearch.asp?search=Selegiline+>

Antidote Dose: The dose to be administered will be specified by the Occ. Med Physician, and is dependent on employee health status.

The University of Minnesota Occupational Health Program is located at:
Boynton Health Service
(612) 626-5422

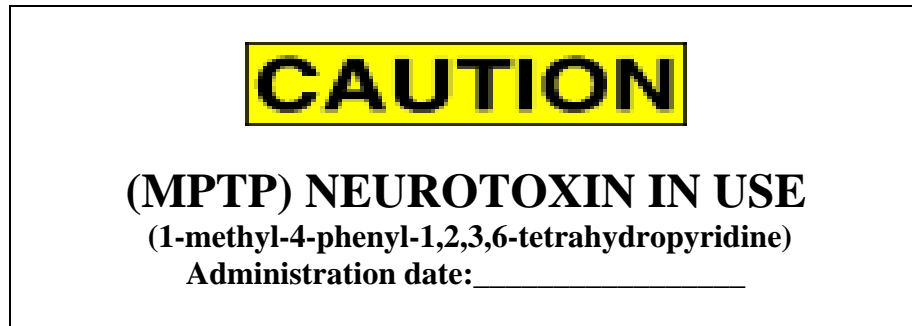
RESEARCH-SPECIFIC STANDARD OPERATING PROCEDURE (SOP)

Antidote must be must be located in the Laboratory where work with MPTP is conducted and must be immediately accessible to all persons who work with MPTP, MPTP solutions or MPTP contaminated materials or specimen

C. Laboratory Doors and Other Work area Signage

- When MPTP is in use, warning signs (see samples below) must be posted on the laboratory door, and the chemical hood until the MPTP has been returned to storage and the work area has been decontaminated.
- A sign must be posted on animal room door/s and on animal cages that contain animals dosed with MPTP (see Animal Safety Protocol).
- Signs will include the information shown below.

Animal Cage Caution Sign



Chemical Hood Sign



RESEARCH-SPECIFIC STANDARD OPERATING PROCEDURE (SOP)

Laboratory or Animal Holding Area Door Sign



NEUROTOXIN (MPTP) IN USE
(1-methyl-4-phenyl-1,2,3,6-tetrahydropyridine)

AUTHORIZED PERSONNEL ONLY

Investigator's Name: _____ Phone# _____

In case of emergency call Environmental Health and Safety at: 726-7273

Material Safety Data Sheets are available in Room _____

Antidote: Selegiline HCl/deprenyl Location _____

RESEARCH-SPECIFIC STANDARD OPERATING PROCEDURE (SOP)

D. Storage

- Pure MPTP and/or MPTP concentrated solutions must be stored in a locked area/cabinet to prevent unauthorized access. Examples are a locked drawer or cabinet. Laboratory door must be locked when authorized personnel are not present lab.
- Store solutions in appropriate, sealed containers with unbreakable secondary containment (i.e., a bottle or vial within a sealed plastic jar). Label all containers, including secondary containment, with the chemical name and hazard warning.
- An inventory of pure MPTP must be kept, and should include the amounts ordered, stock on hand and amounts discarded or used.
- Antidote Selegiline hydrochloride (CAS # 14611-52-0) must be located/stored in the Laboratory where work with MPTP is conducted and must be immediately accessible to all persons who work with MPTP, MPTP solutions or MPTP contaminated materials or specimen.

E. Prudent Work Practices, MPTP Handling and Solution Preparation

- Never work alone when manipulating MPTP or MPTP contaminated materials, always work in pairs, with a partner who's been trained on this SOP, and has Dr. clearance to work with MPTP.
- Work with MPTP or MPTP contaminated materials must be done during regular business hours only.
- Minimize waste, prepare and use the smallest amount of solution necessary for the procedure only
- Opening of pure MPTP, preparation and administration of solutions must take place inside the fume hood only. The fume hood must be/have been certified within the last 6 months. Airflow into and within the hood should not be excessively turbulent, hood face velocity should be adequate (typically 70-100 lfpm).
- Avoid expelling air from syringes containing MPTP solutions to avoid creation of Aerosols. If expelling air is necessary, it must be done inside the hood and into a sterile dampened gauze or cotton ball.
- After injection, **do not clip, remove, or recap the needle**. Instead, place used syringes directly into a sharps container that has been previously placed in the fume hood. Check the injection site for leaking material and absorb with dampened gauze. Contaminated gauzes must be placed in the designated waste container for disposal as hazardous waste.
- Prior to using the fume hood, airflow must be verified by checking the continuous monitoring device (or magnehelic gauge) installed on the hood, if not a smoke test using a smoke generating tube, or a mechanical or electronic device that indicates air flow should be used.

RESEARCH-SPECIFIC STANDARD OPERATING PROCEDURE (SOP)

- During use, the sash must be lowered to the safe operating height of 18 inches.
- Use disposable labware whenever possible. Reusable labware must be decontaminated with a bleach solution before reuse. Disposable lab-ware should be discarded in a designated hazardous waste collection container within the hood.

F. Personal Protective Equipment

The following minimum personal protective equipment must be worn during work with pure MPTP and its solutions:

- Safety goggles or a face shield.
- Disposable Nitrile gloves (NOT latex). Double-gloving is recommended. Change gloves frequently and when contaminated, punctured or torn. Wash hands immediately after removing gloves.
- A disposable laboratory coat, jumpsuit or other appropriate protective clothing must be used. Lab coat or jumpsuit must be removed and discarded as hazardous waste after working with MPTP, and prior to leaving the lab.
- The use of a NIOSH-approved N-95 disposable respirator is recommended when working with pure MPTP or concentrated solutions. Contact UMD Environmental Health and Safety office 726-7273 for guidance on respirator use, selection, fit-testing and training.

G. Preparation and Decontamination

- a. The fume hood's work surface must be lined with a disposable plastic-backed paper liner (blue pads). The liner must be changed after work or experiment is completed and after a spill.
- b. Weighing should take place in the fume hood. If the fume hood produces vibration that prevents accurate weighing, an alternate method is to tare a closed vessel on a balance outside the hood, add the MPTP within the hood, and reweigh the closed vessel outside the hood. Adding of solvent and any additional dilutions must take place in the fume hood.
- c. After work with MPTP, decontaminate all work surfaces and equipment with a solution of household bleach immediately. The fume hood must be decontaminated before any other work is performed in it.
- d. Any unwanted remaining solutions of MPTP as well as any dry waste (paper towels, gloves) must be disposed as hazardous chemical waste through UMD EHS chemical waste disposal program
- e. Wash hands thoroughly immediately after working with any concentration of MPTP.

RESEARCH-SPECIFIC STANDARD OPERATING PROCEDURE (SOP)

4. Animal Handling and Housing (Also See Separate Animal Safety Protocol)

A. General Procedures

- Animal cages containing treated animals must be labeled with the wording "CAUTION: MPTP/NEUROTOXIN" (SEE LABEL ABOVE)
- Use low-dust bedding only, corn-cob bedding or paper bedding is recommended
- Use disposable cages to reduce time working with contaminated materials during cleaning.
- Treated mice must be kept in the laboratory fume hood for three days after dosing (while they are expected to excrete MPTP and its metabolites).
- All potentially contaminated non-metal surfaces must be decontaminated with a bleach solution. Metal surfaces must be washed with a strong detergent solution. Paper towels or rags used for cleaning must be placed in a designated hazardous waste container.

B. Cage Changing and Waste Disposal

- Wear proper personal protective equipment as specified above (Section 3-F) including a NIOSH-approved N-95 respirator.
- Do not change animal bedding for at least 72 hours after administration of MPTP.
- Cage changing will be performed inside the fume hood and by trained staff only.
- Minimize cage changing as much as possible. Cages and bedding may be changed/disposed after 72 hours or in this case after the mice are euthanized.
- Collect and place the bedding and cage in a designated hazardous waste container.
- Leftover drinking water may be mixed with a bleach solution and then disposed in the sewer. Otherwise place as is in the provided waste container
- Leftover feed can be added to and disposed along with the bedding as hazardous waste.
- Place the entire disposable cage into a waste bag, tie the bag and label as per Animal Safety Protocol accordingly, then place it into a designated waste container.

RESEARCH-SPECIFIC STANDARD OPERATING PROCEDURE (SOP)

5. EMERGENCIES: Call 7273 or 911

A. Spills of MPTP outside of the Chemical Fume Hood

For any spill outside of the fume hood including spills of bedding from the first 72 hour after dosing:

- **DO NOT attempt to clean up large spills of MPTP.** Remove outer contaminated clothing first, remove gloves and respirator last and Leave the room immediately, Call the UMD Environmental Health and Safety Office, describe the incident and request assistance.
- Isolate the area to prevent the spread of contamination and post a spill warning sign on laboratory door.
- Alert all personnel in the immediate area to evacuate.
- If personal Injury occurs, call 911 immediately and request immediate assistance.
- NOTE: Personnel may clean up small spills of MPTP or solutions of MPTP which **occurs inside** of the chemical fume hood.
- Follow UMD Emergency procedure: <http://www.d.umn.edu/ehso/emergencies>
- **If exposure, is suspected (i.e. ingestion, skin absorption, inoculation, inhalation, or contact with mucous membrane), have someone call 911. Remove gloves, wash affected area thoroughly for 15 min, and take prescribed dose of selegiline immediately**

B. Spill of MPTP inside the fume hood:

- Small spills may be decontaminated with an excess of 5% to 10% bleach solution. Notify EHSO 7273 of the spill and the cleanup procedure. Place paper towels or absorbing material on the spilled liquid to cover it, and then spray the towels with the bleach solution. This will prevent back splashing or creation of MPTP contaminated aerosols
- If the spill is large or you are unsure of your ability to thoroughly decontaminate it, call EHSO 7273 and request assistance

C. Accidental Exposure to MPTP

In the event of a recognized percutaneous or mucous membrane (eye, nose or mouth) exposure to MPTP

- Immediately take the prescribed dose of the antidote Selegiline HCl.
- Remove contaminated outer clothing first, outer glove, goggles and respirator respectively in that order.

RESEARCH-SPECIFIC STANDARD OPERATING PROCEDURE (SOP)

- Rinse the affected area with water, using a safety shower or eyewash, as appropriate, for at least 15-minutes
- Notify the supervisor, immediately of the incident. The supervisor, a co-worker, or the victim must contact the University Occupational Health Program to determine what additional steps should be taken.
- If the incident occurs off-hours, or an ambulance is needed because of injury, Call 911 and request immediate and necessary assistance.

6. SIGNATURES

Use the following table to list all personnel who will handle MPTP. Initializing indicates that the staff member has read the UMD Medical School Laboratory Safety Plan and this SOP and understands the hazards and safe work practices as detailed in this SOP.

Name	Employee Id #	Initials

Principal Investigator Name: (Print): _____

Principal Investigator (Signature): _____

Date: _____

RESEARCH-SPECIFIC STANDARD OPERATING PROCEDURE (SOP)

7. Contact Information:

<p>Occupational Health/Medicine St. Luke's Occupational Health 218-249-6822 4702 Grand Avenue Duluth, Minnesota 55807 Monday through Friday 8:00 a.m. - 4:30 p.m.</p> <p>Duluth Clinic Occupational Medicine 218-786-3392 400 East Third Street Duluth, Minnesota 55805 Monday through Friday 8:00 a.m. - 4:30 p.m.</p>	<p>After hours, on weekends or holidays St. Mary's Medical Center/Emergency Care and Trauma Center 218-786-4000 407 East Third Street Duluth, Minnesota 55805 Sunday through Saturday 24-hours a day</p>
<p>UMD Env. Health and Safety Office: Health and Safety: 218-726-7273 Waste Management: 218-726-6764</p>	<p>University–Wide Environmental Health and Safety: General: 612-626-6002 Chemical Hygiene Officer:612-626-2330</p>
<p>Occupational Health and Safety Program Ross Janssen , Director (612) 626-5844</p>	

7. REFERENCES

RESEARCH-SPECIFIC STANDARD OPERATING PROCEDURE (SOP)

“*Working With MPTO or MPTP-Treated Animals*,” NIH Division of Safety, Office of Research Services. Posted on AIHA website at:

<http://www2.umdnj.edu/eohssweb/aiha/technical/animal.htm>

Langston, J. W., et al., Chronic Parkinsonism in Humans Due to a Product of Meperidine-analog Synthesis. *Science* 219, 979-980 (1983). Posted at: <http://opioids.com/mptp/>

[Canadian Centre for Occupational Health and Safety \(CCOHS\)](#) RTCS and CHEMINFO databases, for 1-Methyl-4-phenyl-1,2,3,6-tetrahydropyridine (MPTP) CAS # 28289-54-5.

Recommended safe practices for using the neurotoxin MPTP in animal experiments. Yang S. C., Markey S. P., Bankiewicz K. S., London W. T. and Lunn G. (1988) *Lab. Anim. Sci.* 38, 563–567. Posted at:

<http://www.ncbi.nlm.nih.gov/pubmed/3264039?dopt=Abstract>

[The parkinsonian toxin 1-methyl-4-phenyl-1,2,3,6-tetrahydropyridine](#)

(MPTP): a technical review of its utility and safety , *Journal of Neurochemistry*, Vol. 76, No. 5, 2001 1265-1274.

<http://www3.interscience.wiley.com/cgi-bin/fulltext/118974813/PDFSTART?CRETRY=1&SRETRY=0>

[Protocol for the MPTP mouse model of Parkinson's disease](#), Vernice Jackson-Lewis & Serge Przedborski, *Nature Protocols*, Vol 2 No.1 2007

[Chemical Safety in Animal Care, Use, and Research](#)

Wayne R. Thomann, *ILAR Journal* V44(1) 2003.

http://dels.nas.edu/ilar_n/ilarjournal/44_1/v4401thomann.shtml

SOP for the Use of MPTP in Laboratories and Vivarium, University of Medicine and Dentistry of New Jersey, [Environmental and Occupational Health and Safety Services](#).